REMARKS

This response is filed in reply to the Final Office Action dated November 3, 2006 ("Office Action").

Claims 1-15 and 18-46 are pending. Among them, claims 21-45 have been withdrawn from consideration for covering a non-elected invention. Claims 1-15, 18-20, and 46 are under examination.

Applicants have proposed to amend claims 1 and 11 to more clearly set forth the claimed invention. The recitation of "cells remain intact upon removal" in claim 1 is supported in the specification, e.g., at page 23, lines 21-23. Support for "plurality of types of morphogens" recited in claim 11 appears at, e.g., page 8, line 17 to page 9 line 12.[1] The phrase "fragment" and the phrase "laminin, fibronectin, elastin, and thrombospondin" are supported in the specification, e.g., at page 8, line 22 and page 10, lines 3-6, respectively. Applicants have also proposed to amend claim 10 to remove an alleged indefinite term and to cancel claims 18-20. Applicants propose to rewrite claim 6 in independent form. These proposed amendments would add no new matter.

Applicants have filed herewith a Request for Continued Examination and request that the above-mentioned amendment be entered and this application be reviewed in view of the following remarks.

Rejection under 35 U.S.C. § 112, second paragraph

The Office rejected claim 10 for indefiniteness, alleging that the term "sufficient" in the claim is not defined. See the Office Action at page 3, lines 9-15. In the sole interest of moving this case toward allowance, Applicants have deleted the alleged undefined term and respectfully request that the rejection be withdrawn.

[1] This passage of the specification discloses various types of morphogens, including "growth factors, differentiation factors, and bioactive fragments of the ECM itself." Examples of the disclosed growth factors include at least six different types: vascular endothelial growth factor, fibroblast growth factor, insulin-like growth factor, platelet derived growth factor, transforming growth factor, and epidermal growth factor.

Rejection under 35 U.S.C. § 102

The Office rejected claims 1-5, 10-15, 18-20, and 46 for an alleged lack of novelty on various grounds. Applicants will address each ground below.

I

Claims 1-5, 10-12, 18-20, and 46 have been rejected as being allegedly anticipated by Rieck *et al.*, Experimental Cell Research, 1995, volume 220, pages 36-46 ("Rieck"). See the Office Action at page 3, second paragraph. Applicants respectfully disagree and will discuss independent claims 1 and 11 first.

Claim 1 covers a method of generating a morphogen composition from an extracellular matrix (ECM). The method includes (i) growing cells on a surface in a fluid under conditions and for a time sufficient to enable the cells to form an ECM; (ii) removing living cells from the surface and leaving the ECM on the surface, wherein the cells remain intact upon removal; (iii) stimulating the ECM to release morphogens into the fluid; and (iv) collecting the fluid to form a morphogen composition.

In particular, the method requires removing living cells from the ECM and the cells must be intact upon removal. This is desirable given the use of the morphogen composition for wound healing and tissue reconstruction. See, e.g., page 5, last paragraph, of the specification. It is well known in the art that, when cells undergo an accidental death (i.e., necrosis) or are disrupted, they spill out harmful intracellular contents, such as lysosomal contents, and potent enzymes. These intracellular contents can cause an inflammatory response and injure the surrounding cells or tissues. See Nicotera *et al.*, Journal of Cerebral Blood Flow & Metabolism (1999) 19, 583–591. A copy of the abstract of Nicotera *et al.* is attached as Exhibit A. Thus, to prepare a morphogen composition that can be used for tissue reconstruction and wound healing, it is desirable that cells are intact upon removal from an ECM so as not to leave any harmful intracellular contents or cell debris on the ECM.

According to the Office Action, Rieck describes a method of extracting fibroblastic growth factor 2 (FGF2) from an ECM. See the Office Action at page 4, lines 5-6. As the Examiner correctly pointed out, the Rieck method requires "dissolving the cell layer," which is on the ECM, using a detergent Triton-X100. See Rieck, page 37, column 2, lines 1-2, and the Office Action at page 4, line 6. It follows that cells in the cell layers must be already disrupted

by the detergent and undergo necrosis when they are removed from the ECM. As a result, harmful intracellular contents or debris is unavoidably released onto the ECM. Clearly, the Rieck method, which involves "dissolving" cells and removing dead cells from an ECM, differs from that of claim 1, which requires removal of living cells intact from an ECM. Thus, claim 1 is not anticipated by Rieck. .

Claim 11, as amended, covers a morphogen composition comprising a fragment of a molecule selected from the group consisting of laminin, fibronectin, elastin, and thrombospondin. Rieck does not mention laminin, fibronectin, elastin, or thrombospondin, much less a composition comprising a fragment of these molecules, as recited in claim 11. Thus claim 11 is novel over Rieck.

In view of the above remarks and amendments, Applicants submit claims 1 and 11 are novel over Rieck. Thus, claims 2-5, 10, 12-15, 18-20, and 46, all of which depend from claim 1 or 11, are also novel.

II

The Office has rejected claims 11-13, 15, 18-20, and 46 as being allegedly anticipated by U.S. Patent No. 5,714,458 to Adami *et al.* ("Adami"). See the Office Action at page 5, lines 8-9. According to the Office Action, "Adami teaches a stable lyophilized formulation of a fibroblast growth factor (FGF)... which is deemed to be structurally the same as that claimed" in claim 11. See page 5, lines 13-18. Applicants have amended claim 11. The claim, as amended, covers a morphogen composition comprising (1) a plurality types of morphogens and (2) a fragment of a molecule selected from the group consisting of laminin, fibronectin, elastin, and thrombospondin. Adami does not describe these two features. Thus, it does not anticipate amended claim 11, or any claims depending from claim 11, including claims 12-13, 15, 18-20, and 46.

Claims 11-15, 18-20, and 46 have been rejected as being allegedly anticipated by U.S. Patent Application 20020032153 by Whitehouse ("Whitehouse"). See the Office Action at page 6, lines 1-2. Applicants note that Whitehouse does not mention a fragment of a molecule selected from the group consisting of laminin, fibronectin, elastin, and thrombospondin, let alone a composition including this fragment as recited in amended claim 11. Thus, claim 11 and its dependent claims 12-15, 18-20, and 46 are all novel over Whitehouse.

Rejection under 35 U.S.C. § 103(a)

The Office rejected claims 6-9 as being allegedly obvious over Rieck in view of Koyama *et al.*, Nature Biotechnology 1997 ("Koyama") and U.S. Patent Application 20020090725 by Simpson *et al.* ("Simpson"). See the Office Action at page 7, lines 9-12.

Applicants have rewritten claim 6 in independent form and will discuss this claim first. Claim 6 covers a method of generating a morphogen composition from an extracellular matrix. The method includes growing cells on a surface in a fluid under conditions and for a time sufficient to enable the cells to form an extracellular matrix (ECM); removing living cells from the surface and leaving the ECM on the surface, wherein the cells remain intact upon removal; applying an electric potential to the extracellular matrix to release morphogens into the fluid; and collecting the fluid to form a morphogen composition.

Applicants have discussed Rieck above. Koyama describes a controlled culture system, in which astroglial cells are directly attached to and grown on a potential-controlled electrode. A small potential is applied to the cells to induce nerve growth factor (NGF) production. See page 164, left column, paragraph 3.

It is the Office's position that

> one of ordinary skill in the art would have been motivated to use an electric potential to stimulate the secretion of growth factors in the ECM in the method of Rieck because Koyama teaches that electrical stimulation promotes growth factor secretion from cultured cells (which include an ECM) and also because Rieck shows that there is more than one way to extract growth factor from an extracellular matrix. The modulation of the electric potential to comprise varying frequency, potential range, potential cycle shape or potential cycle number would have been a matter of routine optimization for one of ordinary skill in the art. The artisan recognizing that the optimum electric potential cycle and voltage would produce the greatest amount of cell growth and growth factor secretion. (See the Office Action at page 8, lines 9-18)

Applicants would like to point out that claim 6, like claim 1, requires removing living cells intact from an ECM. After the removing step, an electric potential is applied to the ECM to release morphogens. As discussed above, Rieck describes a method that involves disrupting cells on an ECM using a detergent. It does not teach or suggest removing living cells intact from

an ECM, as required in claim 6. Koyama does not cure this defect of Rieck. Accordingly, the two references, alone or combined, do not provide any motivation to remove living cells intact from an ECM, as required in claim 6, and therefore do not render claim 6 obvious.

Further, Koyama's focus is using electric potential to induce the astroglial cells growing on the electrode to produce and secret NGF. Given this focus, one skilled in the art, at best, would have been motivated to apply a potential to living cells, which could produce and secret NGF. He or she would have had no motivation to apply a potential to an ECM from which cells had been removed, because no cells were left to be induced to produce NGF. Thus, to the extent that Koyama focuses on inducing living cells to produce NGF, it teaches away from the method of claim 6.

It is also the Office's position that "[o]ne of ordinary skill in the art would have had a reasonable expectation of success [of combining Rieck and Koyama's teachings to obtain morphogens from an ECM] because Simpson teaches that an electrical field can stimulate movement or conformational changes in a matrix due to the movement of magnetically or electrically sensitive particles." See the Office Action at page 2, line 6, through page 4, line 11. Applicants respectfully disagree.

Simpson describes electrospinning, electrospraying, electroaerosoling, or electro-sputtering of collagen toward a target so as to form an implantable scaffold. Bioactive materials, such as growth factors, can be incorporated into the scaffold in the same process. The resulting loaded scaffold can be implanted into a tissue where the bioactive materials are released. See, e.g., paragraphs 0007, 0009, 0012, and 0046. In view of these descriptions, it is clear that the Simpson method focuses on incorporating bioactive materials into a collagen matrix (which can be used as a vehicle to deliver the bioactive materials into a tissue). This is the opposite of the method of claim 6, which obtains a morphogen from an ECM. Thus, one skilled in the art would not have had a reasonable expectation of success in combining Simpson with Rieck or Koyama to obtain morphogens from an ECM.

Furthermore, the man-made collagen matrix described in Simpson is quite different from an ECM as recited in claim 6. It is known in the art that an ECM is a complex network of a large number of polysaccharides and different proteins secreted by cells. It is far more complex than the man-made collagen matrix described in Simpson. In view of this complexity, one skilled in

the art would not have had a reasonable expectation of success of using the Simpson method for man-made collagen matrix to induce morphogens to be released from the ECM described in Rieck (which, as noted above, is a contaminated ECM that is very different form the ECM recited in the ECM recited in the Applicants' claimed method).

In view of the above remarks, Applicants submit that claim 6 is non-obvious over Rieck in view Koyama and Simpson. Claims 7-9 depend from claim 6. At least for the same reasons set forth above, they are also non-obvious.

CONCLUSION

Applicants submit that grounds for the rejections asserted by the Examiner have been overcome, and that the pending claims define subject matter that is patentable. As a result, applicants submit that allowance of this application is proper, and request an early favorable action.

Enclosed is a Request for Continued Examination and a Petition for One Month Extension of Time. Fees in the amount of $395 and $60 are being paid concurrently on the Electronic Filing System (EFS) by way of Deposit Account authorization. Please apply any other required fees to Deposit Account No. 06-1050, referencing the attorney docket number shown above.

Respectfully submitted,

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Exhibit A

1: J Cereb Blood Flow Metab. 1999 Jun;19(6):583-91.

Excitotoxins in neuronal apoptosis and necrosis.

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Neuronal loss is common to many neurodegenerative diseases. Although necrosis is a common histopathologic feature observed in neuropathologic conditions, evidence is increasing that apoptosis can significantly contribute to neuronal demise. The prevalence of either type of cell death, apoptosis or necrosis, and the relevance for the progression of disease is still unclear. The debate on the occurrence and prevalence of one or the other type of death in pathologic conditions such as stroke or neurotoxic injury may in part be resolved by the proposal that different types of cell death within a tissue reflect either partial or complete execution of a common death program. Apoptosis is an active process of cell destruction, characterized morphologically by cell shrinkage, chromatin aggregation with extensive genomic fragmentation, and nuclear pyknosis. In contrast, necrosis is characterized by cell swelling, linked to rapid energy loss, and generalized disruption of ionic and internal homeostasis. This swiftly leads to membrane lysis, release of intracellular constituents that evoke a local inflammatory reaction, edema, and injury to the surrounding tissue. During the past few years, our laboratories have studied the signals and mechanisms responsible for induction or prevention of apoptosis/necrosis in neuronal injury and this is the subject of this review.

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